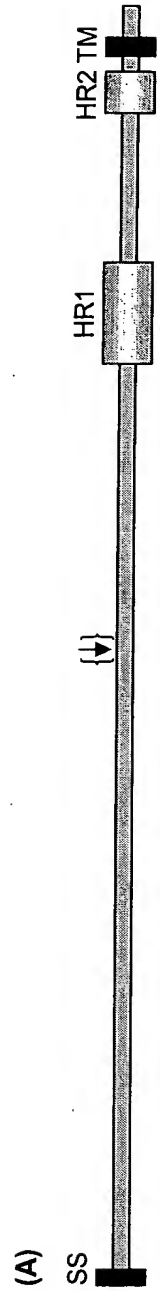
TITLE: CORONA-VIRUS-LIKE PARTICLES COMPRISING FUNCTIONALLY DELETED GENOMES
Inventor: Rottier et al.
Docket No.: 2183-6265US

FIG.1



(B)

HR1 region:

```
                Frame I  d  a  d  a  d  a
```

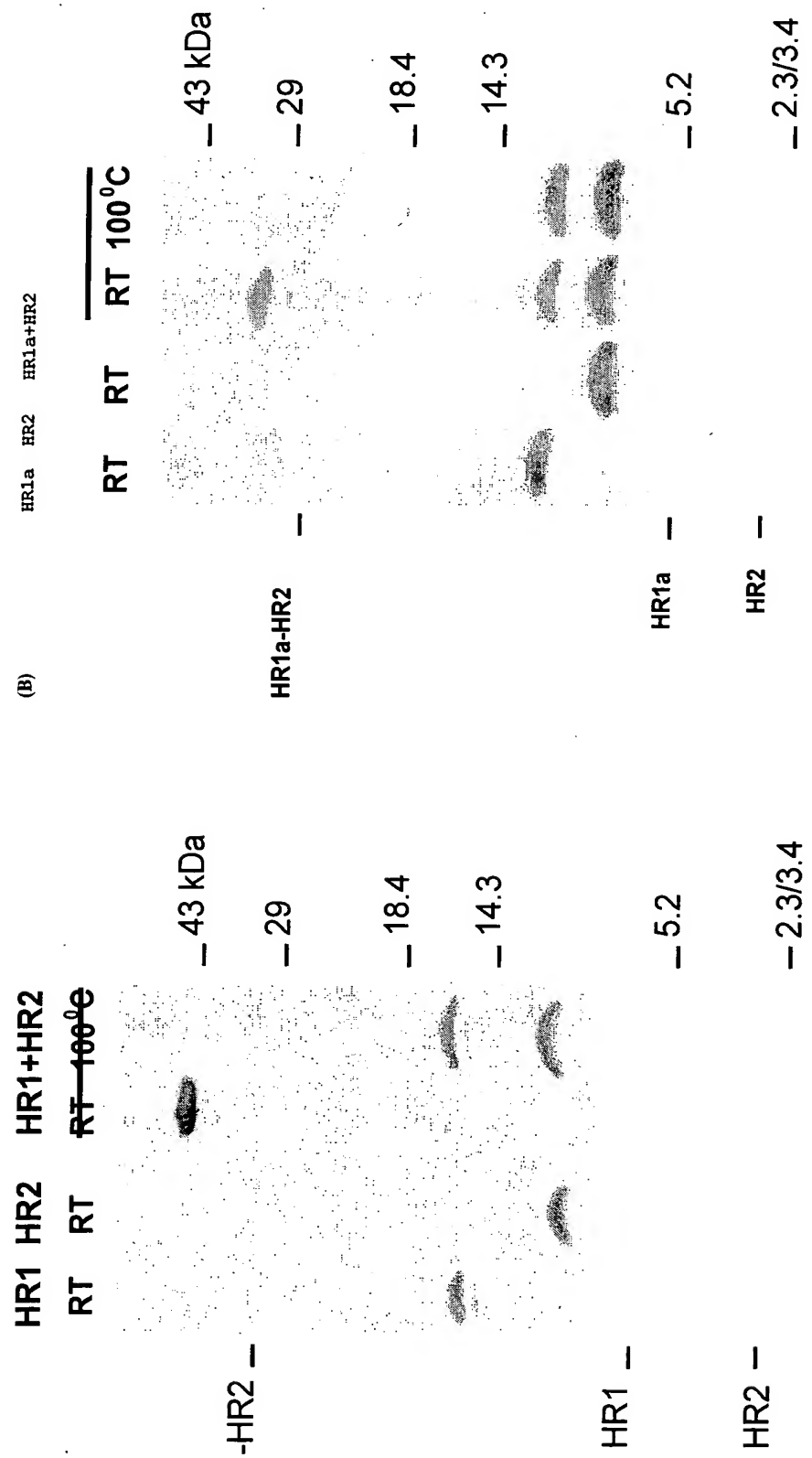

(A)
HR1 HR2 HR1+HR2
RT RT RT 100°C
43 kDa
29
18.4
14.3
5.2
2.3/3.4
HR1-HR2
HR1
HR2
(B)
HR1a HR2 HR1a+HR2
RT RT RT 100°C
43 kDa
29
18.4
14.3
5.2
2.3/3.4
HR1a-HR2
HR1a
HR2

Fig. 2

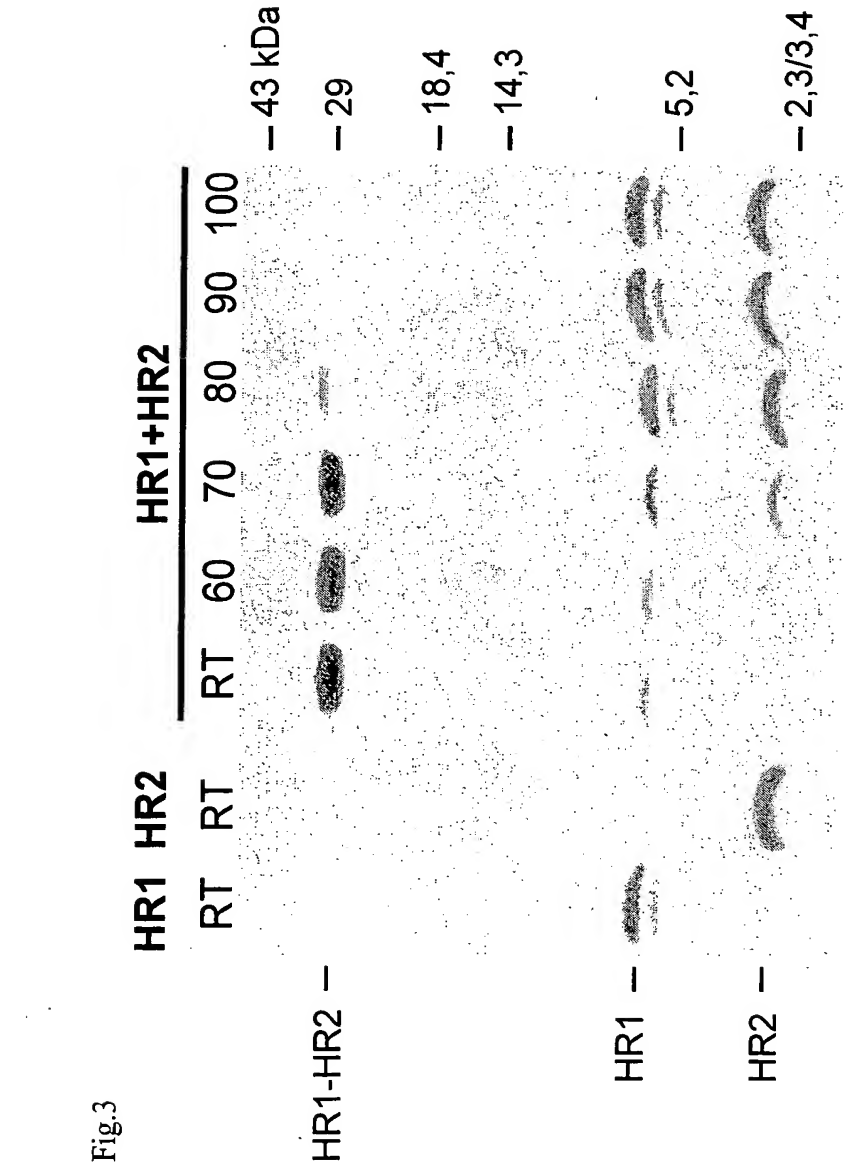

HR1 HR2
HR1+HR2
RT RT RT 60 70 80 90 100
43 kDa
29
18,4
14,3
5,2
2,3/3,4
HR1-HR2
HR1
HR2

Fig.3

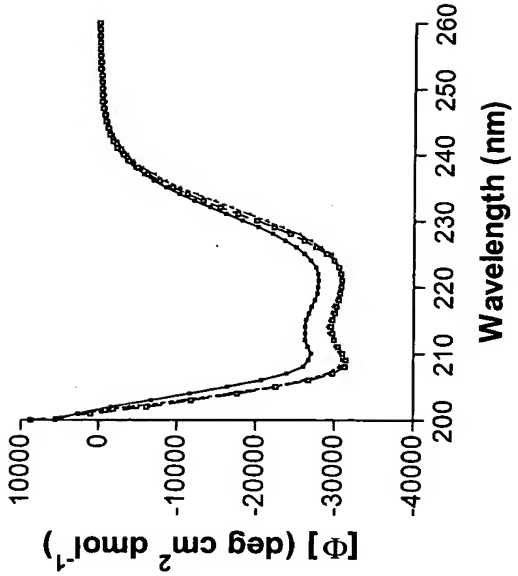

10000
0
-10000
-20000
-30000
-40000
200
210
220
230
240
250
260
Wavelength (nm)
[Φ] (deg cm² dmol⁻¹)

Fig. 4.

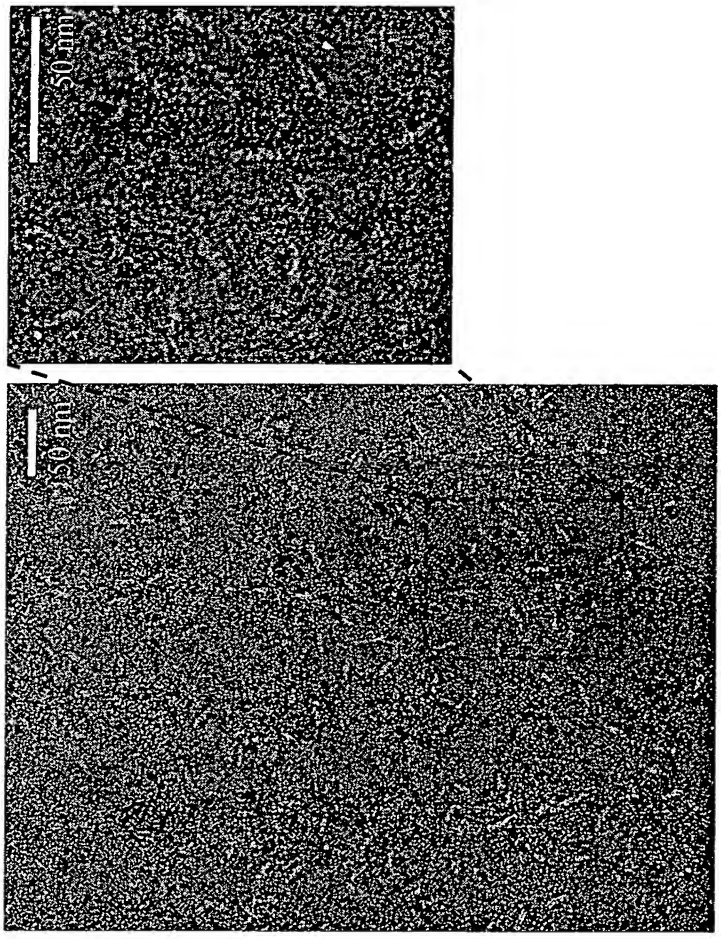


Fig. 5

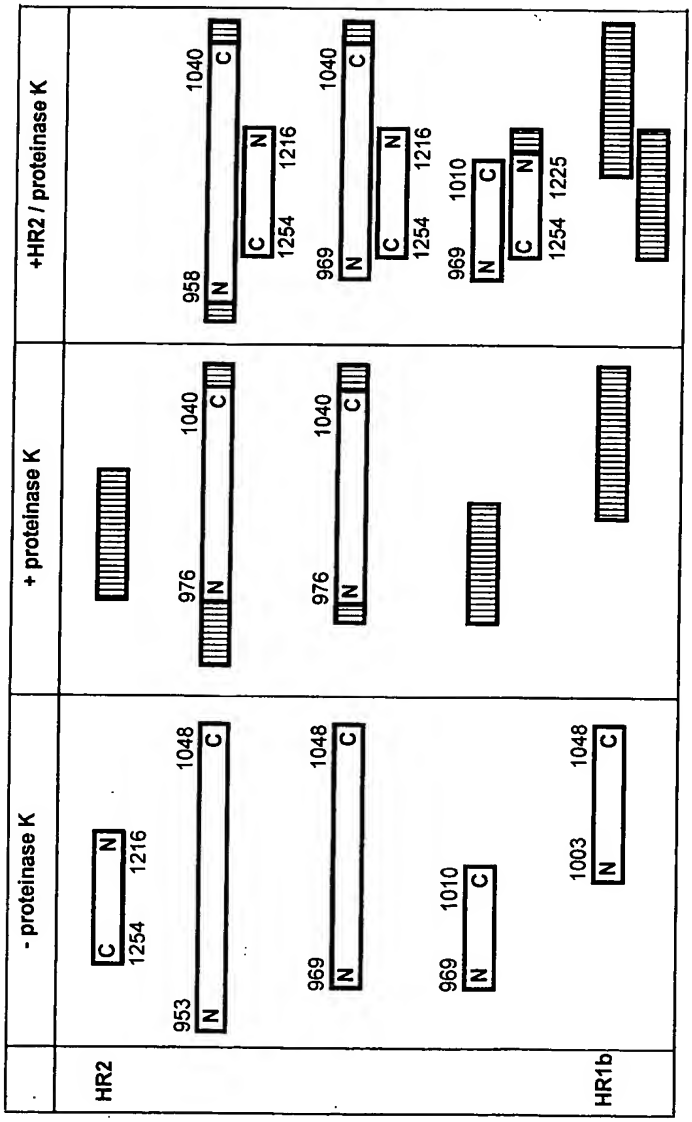
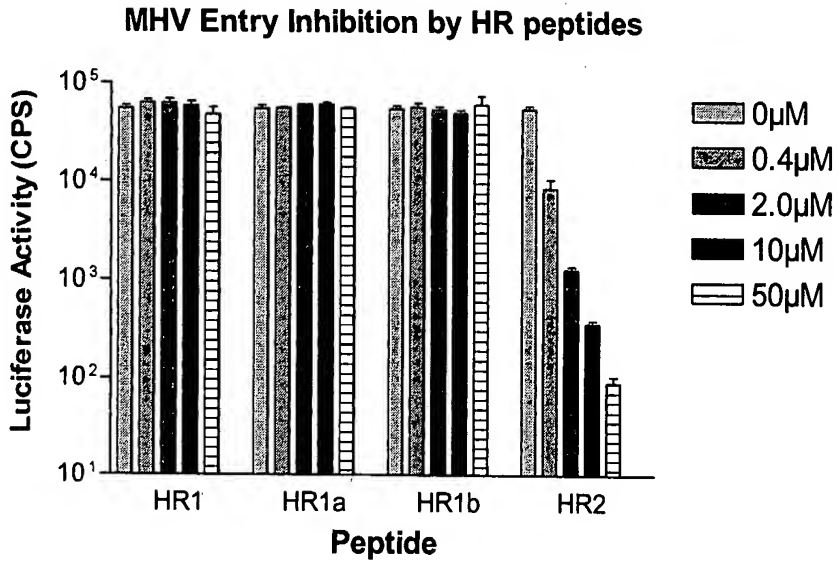

- proteinase K
+ proteinase K
+HR2 / proteinase K
HR2
HR1b
N
C
953
969
976
1003
1010
1040
1048
1216
1225
1254
958

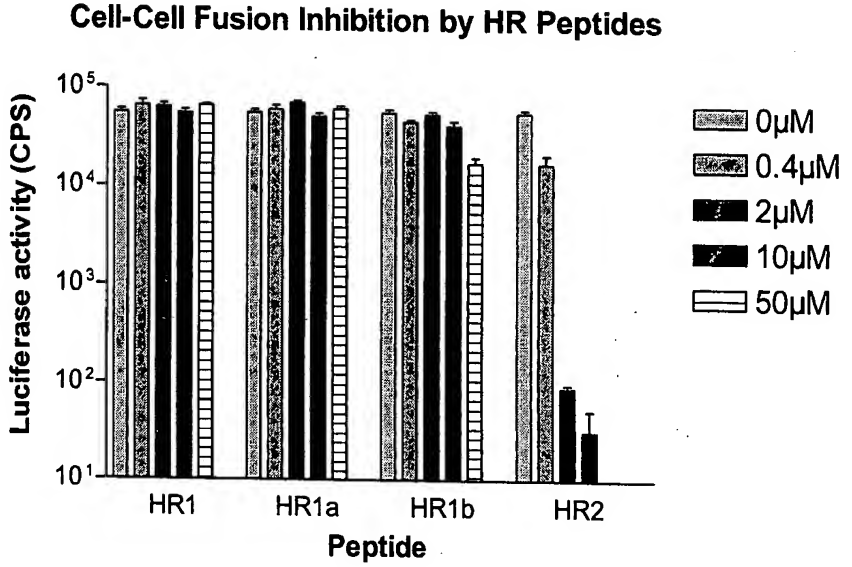
Fig.6

Fig.7

(A)



(B)



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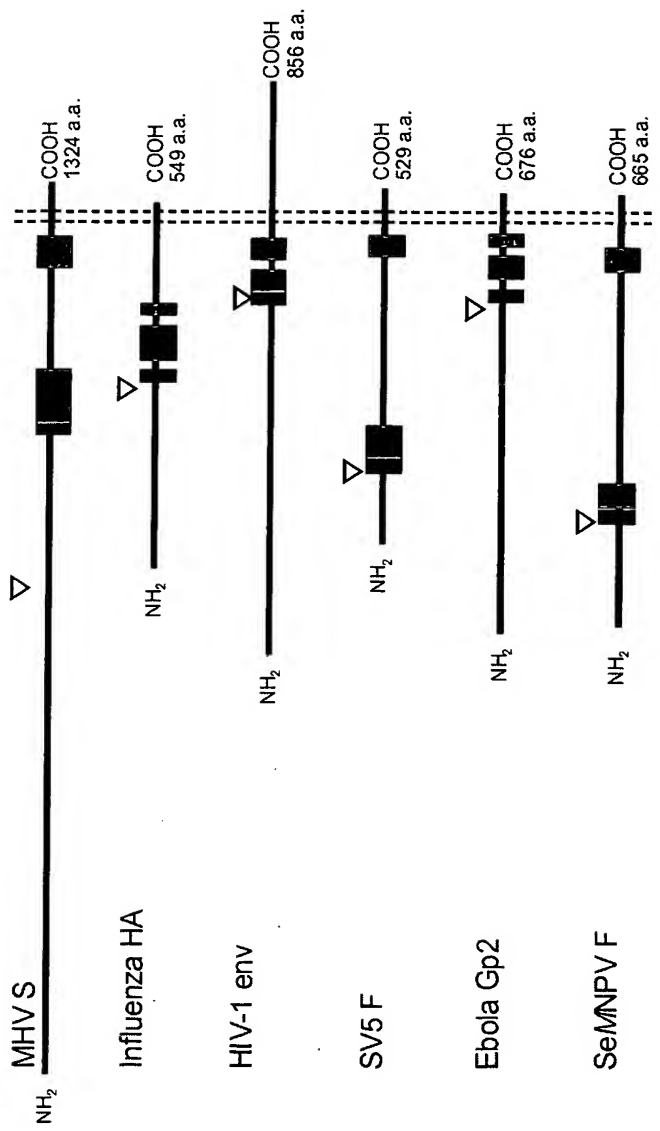

Fig 8

Fig.9

>GST-**HR1**
MSPILGYWKIKGLVQPTRLLLEYLEEKYEEHLYERDEGDKWRNKKFELGLEFPNLPYYIDGDVKLTQSMAIIRYIADKHNMLGGCPKERAEISMLEGAVLDIRYGVSRIAYSKDFE
TLKVDFLSKLPEMLKMFEDRLCHKTYLNGDHVTHPDFMLYDALDVVLYMDPMCLDAFPKLVCFKKRIEAIPQIDKYLKSSKYIAWPLQGWQATFGGGDHPPKSDLVPRGS**QARLNY**
VALQTDVLNKNQQILANAFNQAIGNITQAFGKVNDAIHQTSQGLATVAKALAKVQDVVNTQGQALSHLTVQLQNNFQAISSSISDIYNRLDELSADAQVDRLIT

>GST-**HR2**
MSPILGYWKIKGLVQPTRLLLEYLEEKYEEHLYERDEGDKWRNKKFELGLEFPNLPYYIDGDVKLTQSMAIIRYIADKHNMLGGCPKERAEISMLEGAVLDIRYGVSRIAYSKDFE
TLKVDFLSKLPEMLKMFEDRLCHKTYLNGDHVTHPDFMLYDALDVVLYMDPMCLDAFPKLVCFKKRIEAIPQIDKYLKSSKYIAWPLQGWQATFGGGDHPPKSDLVPRGS**EFTLDI**
FNATYLNLTGEIDDLEFRSEKLHNTTVELAILIDNINNTLVNLEW

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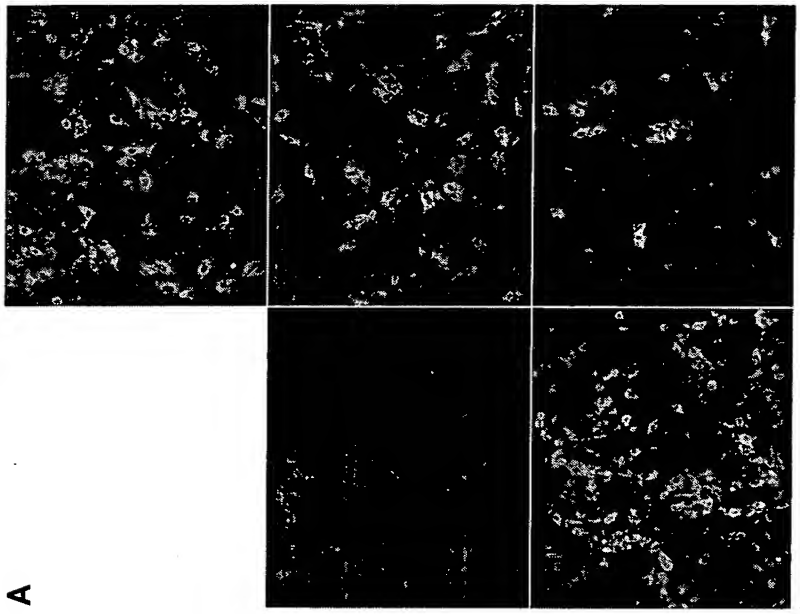
Fig. 10

>SARS nucleotide and deduced protein sequence as derived from the RT-PCR fragment:

```
nt 1 gaaatchcgscttctgctaatcttgctgctactaaaatgtctgagtgtgttcttggacaa
aa 1  E  I  X  X  S  A  N  L  A  A  T  K  M  S  E  C  V  L  G  Q
tcaaaaagagttgacttttgtggaaagggctaccaccttatgtccttcccacaagcagcc
 S  K  R  V  D  F  C  G  K  G  Y  H  L  M  S  F  P  Q  A  A
ccgcatggtgttgtcttcctacatgtcacgtatgtgccatcccaggagaggaacttcacc
 P  H  G  V  V  F  L  H  V  T  Y  V  P  S  Q  E  R  N  F  T
acagcgccagcaatttgtcatgaaggcaaagcatacttccctcgtgaaggtgtttttgtg
 T  A  P  A  I  C  H  E  G  K  A  Y  F  P  R  E  G  V  F  V
tttaatggcacttcttggtttattacacagaggaacttcttttctccacaaataattact
 F  N  G  T  S  W  F  I  T  Q  R  N  F  F  S  P  Q  I  I  T
acagacaatacatttgtctcaggaaattgtgatgtcgttattggcatcattaacaacaca
 T  D  N  T  F  V  S  G  N  C  D  V  V  I  G  I  I  N  N  T
gtttatgatcctctgcaacctgagcttgactcattcaaagaagagctggacaagtacttc
 V  Y  D  P  L  Q  P  E  L  D  S  F  K  E  E  L  D  K  Y  F
aaaaatcatacatcaccagatgttgatcttggcgacatttcaggcattaacgcttctgtc
 K  N  H  T  S  P  D  V  D  L  G  D  I  S  G  I  N  A  S  V
gtcaacattcaaaaagaaattgaccgcctcaatgaggtcgctaaaaatttaaatgaatca
 V  N  I  Q  K  E  I  D  R  L  N  E  V  A  K  N  L  N  E  S
ctcattgaccttcaagaattgggaaaatatgagcaatatatt    582
 L  I  D  L  Q  E  L  G  K  Y  E  Q  Y  I     194
```

FIG.11

A



B

peptide	amino acid sequence	EC50 ± SD (μM) SCV	MHV
B.1			
sHR2-1	ELDSFKEELDKYFKNHTSPDVDLGDISGINASVVNIQKEIDRLNEVAKNLNESLIDLQELGKYE	4336.4	>50
sHR2-2	FKEELDKYFKNHTSPDVDLGDISGINASVVNIQKEIDRLNEVAKNLNESLIDLQELGKYE	2832.8	nd
sHR2-3	LDKYFKNHTSPDVDLGDISGINASVVNIQKEIDRLNEVAKNLNESLIDLQELGKYE	>50	nd
sHR2-4	FKNHTSPDVDLGDISGINASVVNIQKEIDRLNEVAKNLNESLIDLQELGKYE	>50	nd
sHR2-5	TSPDVDLGDISGINASVVNIQKEIDRLNEVAKNLNESLIDLQELGKYE	>50	nd
sHR2-6	VDLGDISGINASVVNIQKEIDRLNEVAKNLNESLIDLQELGKYE	>50	nd
sHR2-7	DISGINASVVNIQKEIDRLNEVAKNLNESLIDLQELGKYE	>50	nd
sHR2-8	ELDSFKEELDKYFKNFSPDVDLGDISGINASVVNIQKEIDRLNEVAKNLNESLIDLQELGKYEQYIK	1723.0	nd
sHR2-9	ELDSFKEELDKYFKNFSPDVDLGDISGINASVVNIQKEIDRLNEVAKNLNESLIDLQEL	3434.0	nd
sHR2-10	ELDSFKEELDKYFKNTSPDVDLGDISGINASVVNIQKEIDRLNEVAKNLNESLID	>50	nd
mHR2	DLSIDFEKLNVYLLDLTYEMNRIQDAIKKLNESYINLKE	>50	0.920.1
B.2			
sHR1	AYRFNGIGVTQNVLYENQKQIANQFNKAISQIQESLTTTSTALGKLQDVVNQNAQALNTLVKQ-	>50	nd
sHR1a	NQKQIANQFNKAISQIQESLTTTSTALGKLQDVVNQNAQALNTLVKQ-	>50	nd
sHR1b	NQNAQALNTLVKQLSSNFGAISSVLNDILSRLDKVEAEVQIDRLIT	>50	nd
sHR1c	NQKQIANQFNKAISQIQESLTTSTALGKLQDVVNQNAQALNTLVKQ	>50	nd

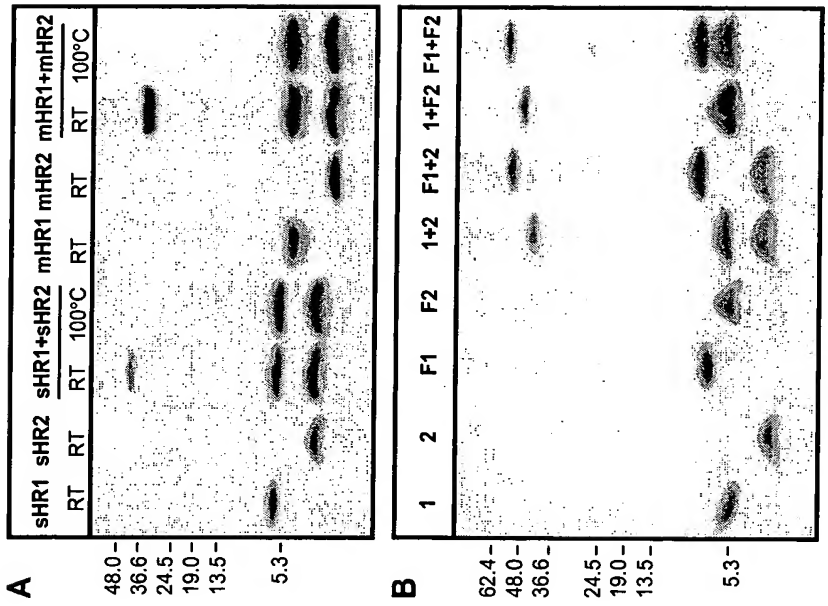

A
sHR1 sHR2 sHR1+sHR2 mHR1 mHR2 mHR1+mHR2
RT RT RT 100°C RT RT RT 100°C
48.0
36.6
24.5
19.0
13.5
5.3
B
1 2 F1 F2 1+2 F1+2 1+F2 F1+F2
62.4
48.0
36.6
24.5
19.0
13.5
5.3

FIG.12

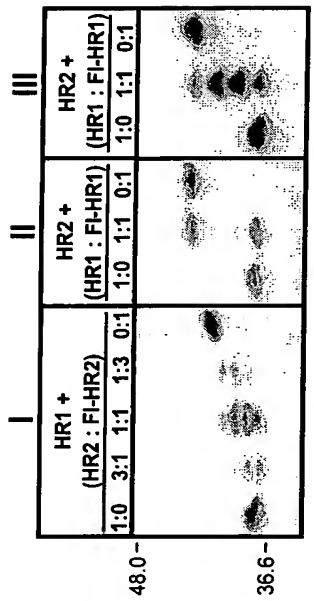

I
II
III
HR1 +
(HR2 : FI-HR2)
1:0 3:1 1:1 1:3 0:1
HR2 +
(HR1 : FI-HR1)
1:0 1:1 0:1
HR2 +
(HR1 : FI-HR1)
1:0 1:1 0:1
48.0
36.6

FIG.13

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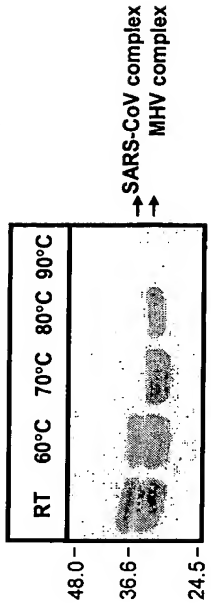

FIG.14

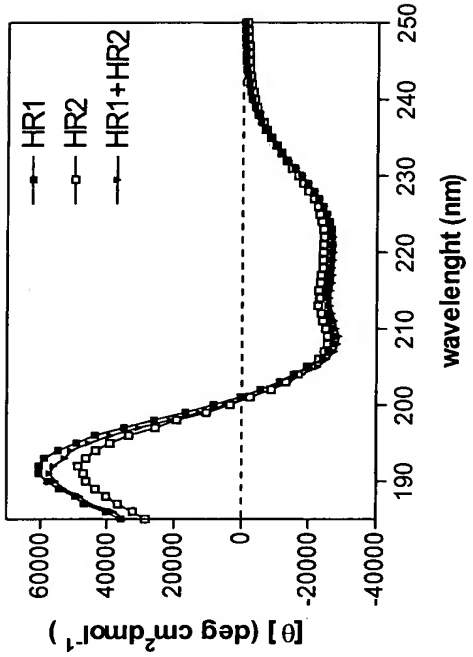
HR1
HR2
HR1+HR2
60000
40000
20000
0
-20000
-40000
190
200
210
220
230
240
250
wavelenght (nm)
[θ] (deg cm²dmol⁻¹)

FIG.15

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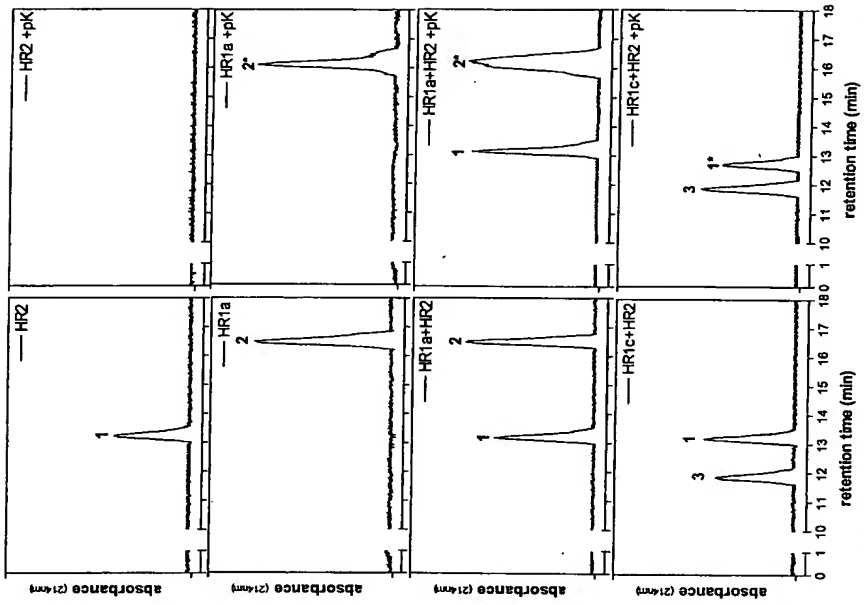

Peak nr.:	Amino acid sequence:	Ma Mw:	Pred. Mw:
1 (HR2)	GSDVDLGDISGINASVVNIQKEIDRLNEVAKNLNESLIDLQELGKYE	5129	5130
1*	GINASVVNIQKEIDRLNEVAKNLNESLIDLQELGKYE	4170	4171
2 (HR1a)	GSNQK--LNTLVKQLSSNFGAISSVLNDILSRLDKVEAEVQIDRLIT	8884	8885
2*	GSNQK--LNTLVKQLSSNFGAISSVLNDILSRLDKVEAEVQIDRLIT	8884	8885
	GSNQK--LNTLVKQLSSNFGAISSVLNDILSRLDKVEAEVQIDRL	8669	8670
	GSNQK--LNTLVKQLSSNFGAISSVLNDILSRLDKVEAEVQID	8400	8400
	GSNQK--LNTLVKQLSSNFGAISSVLNDILSRLDKVEAEVQ	8172	8173
3 (HR1c)	GSNQK--LNTLVKQ	5285	5286

FIG.16

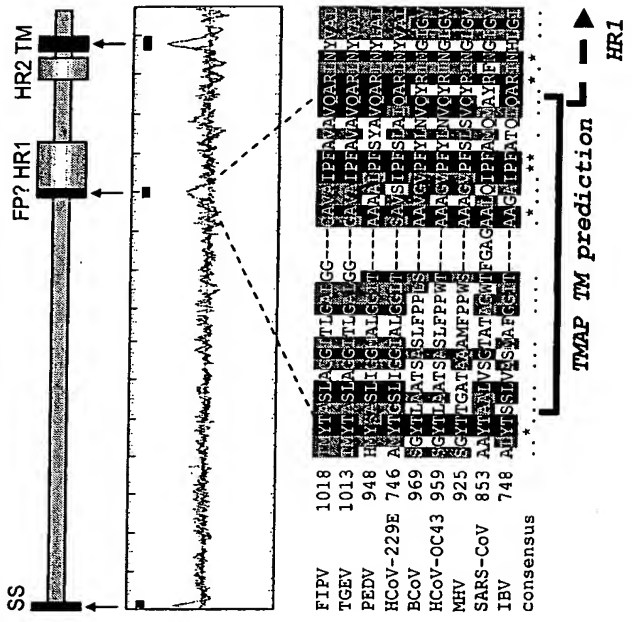
SS
FP? HR1
HR2 TM
FIPV 1018
TGEV 1013
PEDV 948
HCoV-229E 746
BCoV 969
HCoV-OC43 959
MHV 925
SARS-CoV 853
IBV 748
consensus
TMAP TM prediction
HR1

FIG.17